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EXHIBIT A

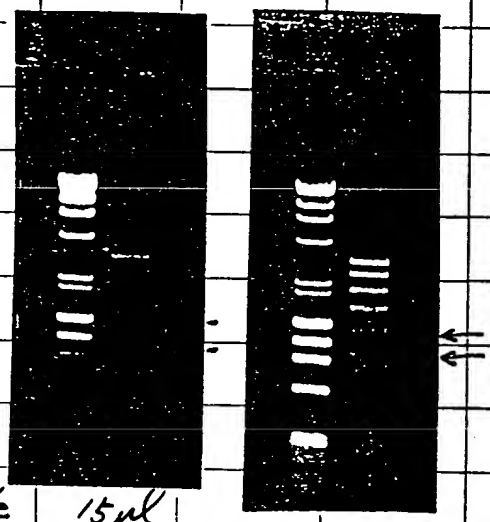
Subcloning p11 (pT1-HP): the Hind III-PstI 0.95kb Fragment of Genomic Clone T11

870409

Subcloning p11 / Sac I + Pst I
Pst I - Hind III

Digestion of p11-6 by Sac I - Hind III - Pst I

DNA	45μl	(≦ 3μg)	1μg p11-6 (4/9 miniprep.)
Sac I	2μl		
10×B	20μl		
ddH2O	133μl		
total	200μl	37°C 1hr	
Hind III	2μl		Sac-Pst I 1.2kb 80ng
10×B	10μl		Pst I - Hind III 0.95kb 63ng
1M Tris 8.0	8μl		
5M NaCl	2μl		
ddH2O	78μl		
total	300μl	37°C 1hr	
Pst I	2μl		
~~10×B~~	~~10μl~~		
5M NaCl	3μl		
ddH2O	95μl		
	400μl	37°C 1hr → check	15μl
	5μl	(5μg tRNA)	
	12μl	(5M NaCl)	
	↓		
	Phenol ext.	5min.	(→ CIAA ext)
	↓ cfg	10min.	
	EtOH 1ml	dry ice.	20min. → cfg 15min
	↓ Wash	~ 70% EtOH	→ cfg 5min
	↓ lyophylize	7min	
	↓ Resolve in [redacted]μl	TE + BT	8μl



32

870409

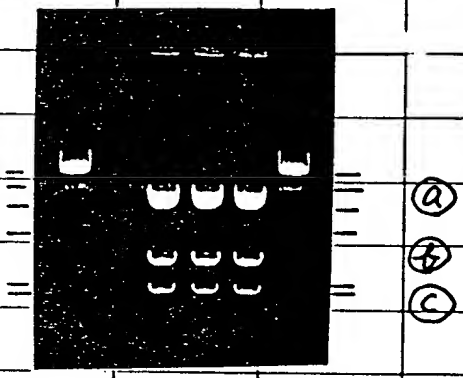
Subcloning pII / SacI - HindIII 2.2kb fragment (cont.)

resalve in 50μl TE Add 5μl B-J

EP 1% Agarose (Embrya gel 3 lanes) λ/H 0.5μg

Gene clean.

ⓒ	Agarose	0.1g	
	NAI	25ml.	250μl
	Glassmilk	5μl	
	NEW		250μl
	TE	2.5μl×2	

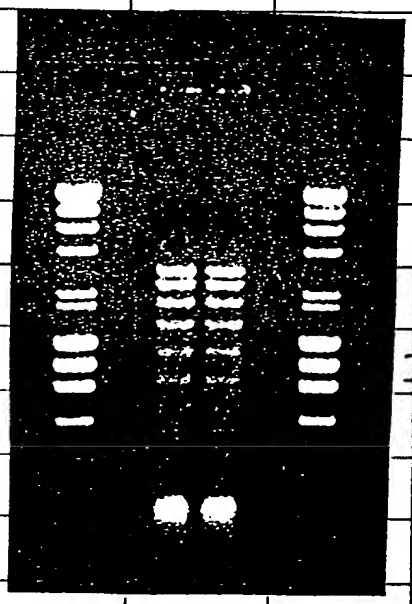


Subcloning pII / SacI - PstI 1.2kb insIV

pII / PstI - HindIII 0.95kb

EP 1% Agarose (Minigel 2 lanes)

Gene clean	1.2	0.95
Agarose	0.07	0.18
NAI	175μl	450
Glassmilk	5μl	5μl
NEW	250μl	250μl
TE	2.5×2	2.5×2



Sequencing p11 (0.95kb)

870412

Sequencing of pII - PstI HindIII /0.95kb

#128-2 18μl. Alkali denature

(left primer (18-mer)
right primer

16000V·hr /1300V = 12.3hr

1 20:45 →
2 22:35 → 10:55

Fixation 10% methanol /10% Acetate /2l
15 min. after detaching gel from plate
Gel dryer 80°C 50min
Autoradiography O/N RT

IFIND - INTELLIGENETICS T· .M. Page 1

v-fms

FIIPHL---FESVSHONC

```
  210       200       190       180       170       160
NCCTTGCAGACCTTTCTGCCCGTGAAGTGGATG--CTCCTGAGAGCATCTTTGACAACCTC
          : ::   ::::: :::::::::::: :::: :::::::::::: :::     ::
AAGGGCAATGCCCGCCTGCCTGTGAAGTGGATGGCTCCGGAGAGCATCTTCGACTGTGTC
          4150      4160      4170      4180      4190

   150       140       130       120       110       100
TACACCACACTGAGTGATGTCTGGTCTTATG--CATTCTGCTCTGGGAGATCTTTTCCCTT
:::::     : ::: :: :::::::: :::: :::  :::::::::: ::::: :: ::
TACACGGTTCAGAGCGACGTCTGGTCCTATGGCATCTTGCTCTGGGAAATCTTCTCACTC
          4210      4220      4230      4240      4250

    90        80
CGTATGGGCCTGACATTGCT
:   ::  ::
CGGCTGAACCCCTACCCTGG
          4270
```

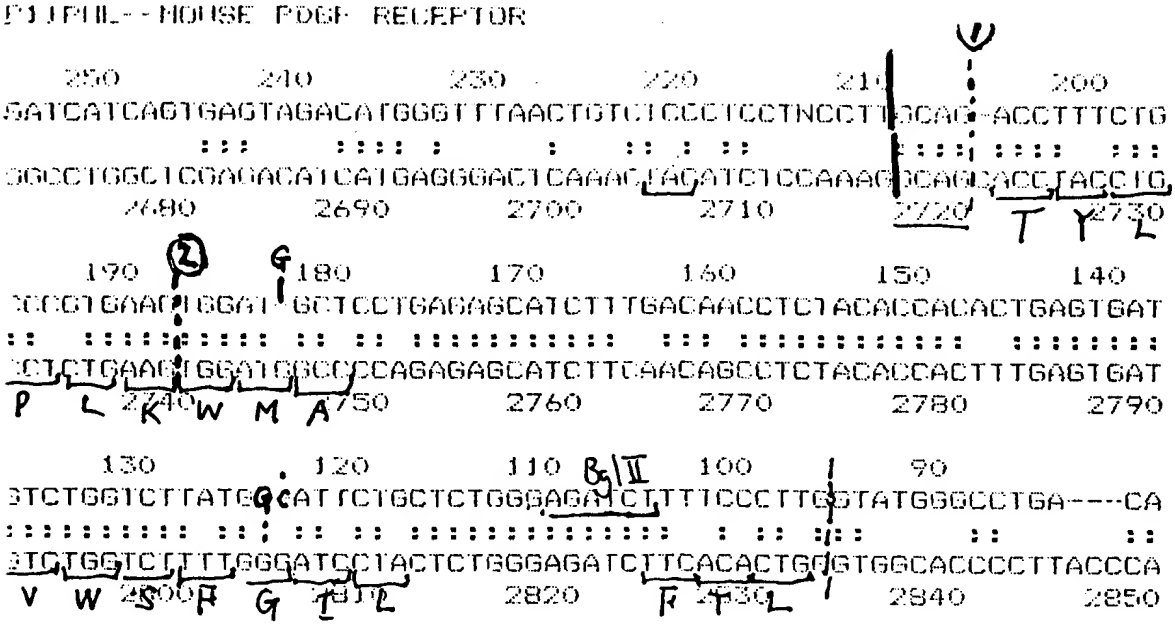
① 88/112 bp = 78.6%

② 74/94 bp = 78.7%

Score=38, Matched=90, Mismatched=28, Unmatched=2, Gaps=2
Window=20, Word-size=2, Density=Less, Gap-Penalty=4

rat-Kc

P1JPHL--MOUSE PDGF RECEPTOR



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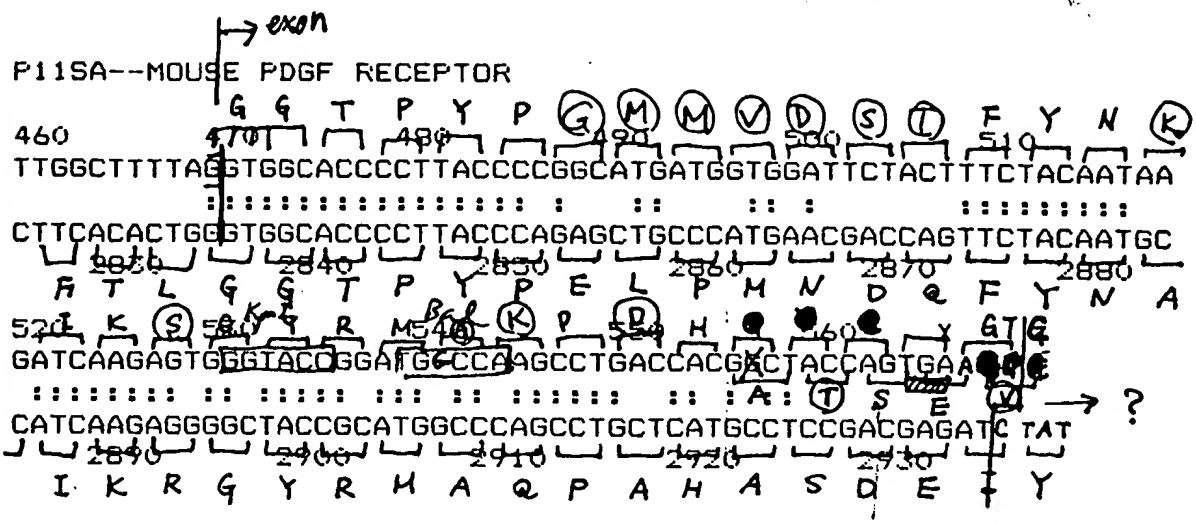
76% c m

50% - h m

① 93/112 = 83.0%

② 79/94 = 84.0%

IFIND - INTELLIGENETICS Page 1



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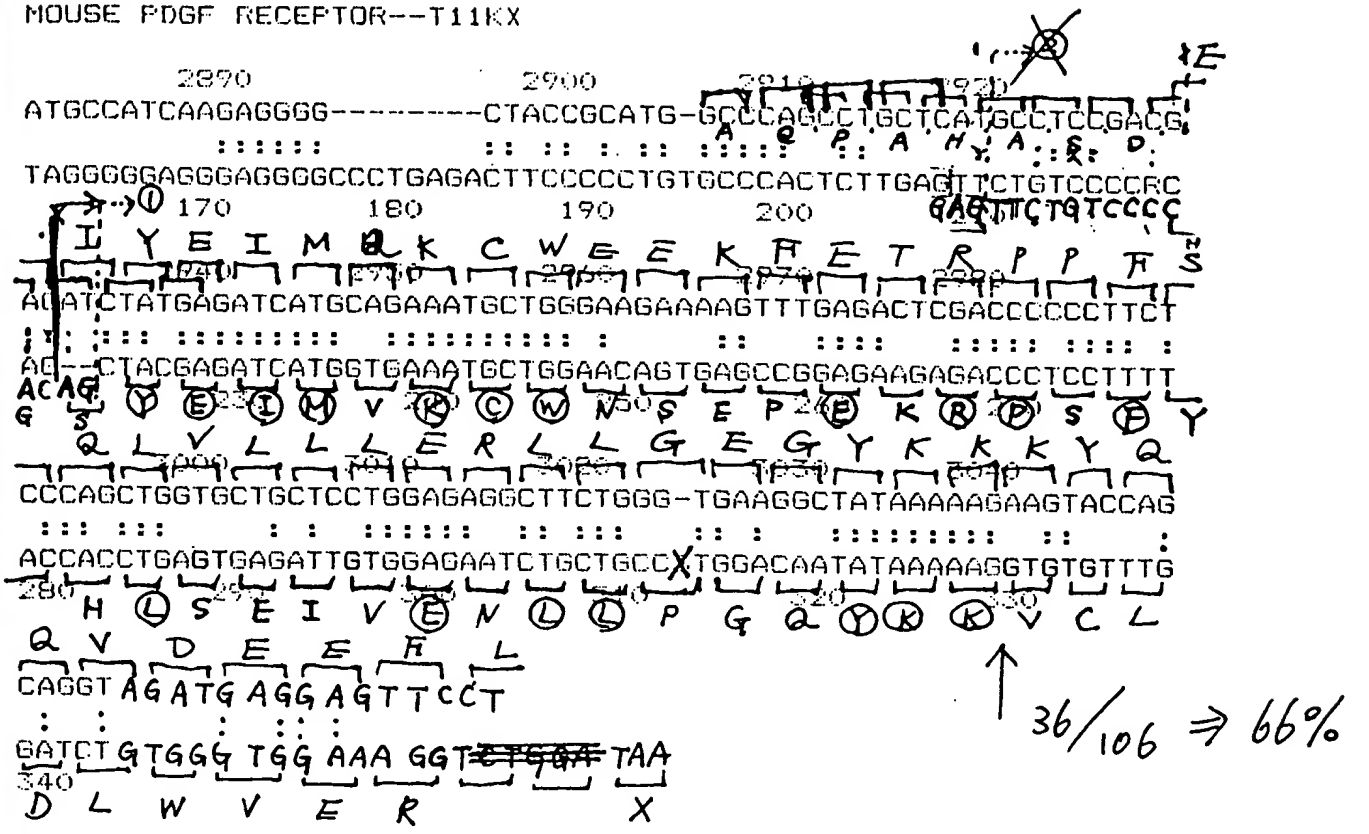
MOUSE PDGF RECEPTOR--T11KX



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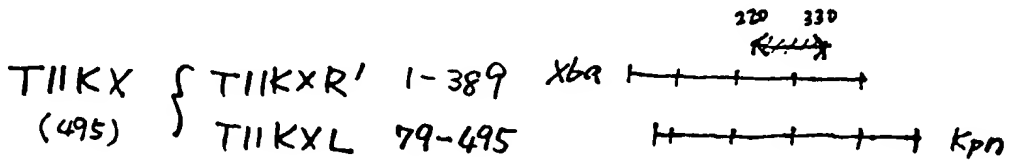
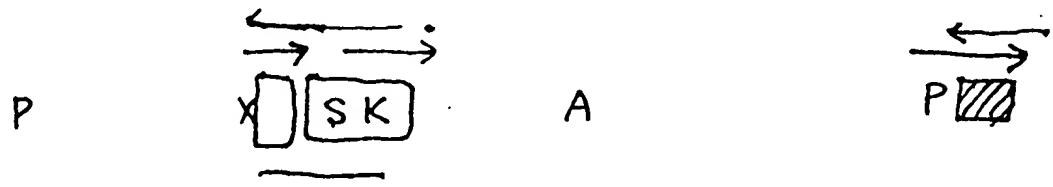




Intron

GT-AG

Titration and Mini-Prep of the Okayama-Berg
cDNA Library (Normal Human Fibroblasts cDNA Library)

870629

Titration of O-B cDNA library (DH5)

9.5×10^4/ml

1×10^6/μl (1×10^9/ml) DH1 1 OD550 = 0.5 (~ 5×10^7 cells/ml

10μl → 1ml 1×10^4/μl

10μl	10^3	4	111
1μl	10^2	3	11
10μl	10	2	
1μl	1	1	

10μl → 1ml 1×10^2/μl

10μl → 1ml 1 /μl

22:46' – 10:45' (12h

– 16:30' (18h

Titer : 1.1×10^5/μl

↓

13.6μl

870630

1.5×10^8 /plate × 66 plate

10μl /plate × 66 plates (660μl) 1.5×10^6 / 1000μl

870710

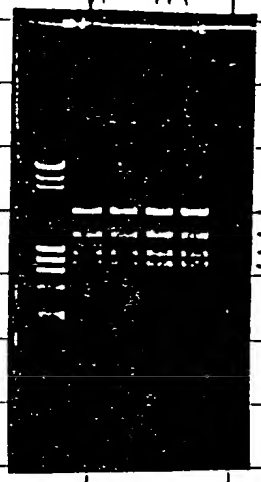
#248

Mini-prep. of 0-B cDNA clone 1

o/N liquid cultures were stored in 15% glycerol at -70° C

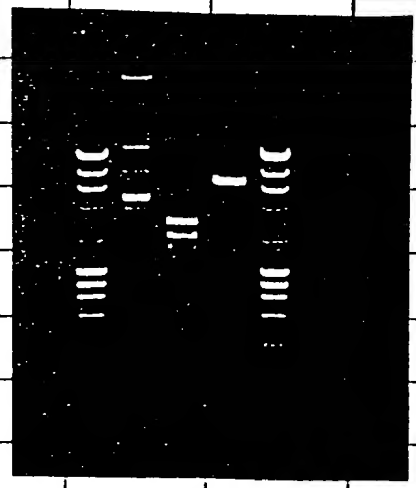
#248

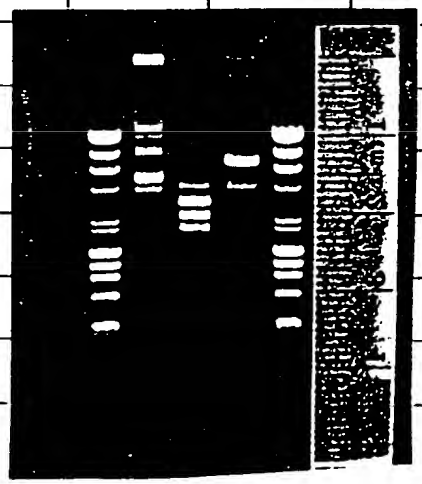
DNA	4 µl	(-1~-4)
BamH I	1 µl	
10×B	1 µl	
ddH2O	4 µl	
Total	10 µl	



DNA	4 µl	(1-1)
Sal I or Xho I	1	
10×B	1	
dd H2O	4	
Total	10 µl	

Cont. Xho I Sal I





870711 M

Isolation of 0.2kbp Subfragment of λEF 17 To Screen a M426 Human Embryo Fibroblast cDNA Library

#420

880103

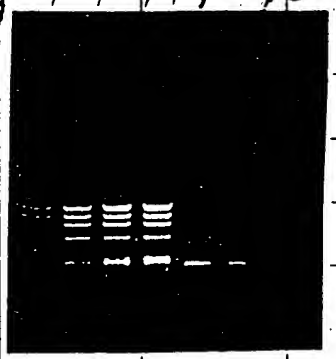
Isolation of EcoRI 0.2kb fragment from pMF17-2

#352 437 ng/μl pUC13

DNA	343μl	150μg	× 0.2 / 2.7+0.2
EcoRI	75μl		
10×B	50μl		
RNase	10μl		
ddH2O	22		
total	500μl		37°C 20min

4% NuSieve gel 15V const. o/n w/o circulation

	EcoRI 0.2	T11	T11DEC
gel	0.83	1.93	1.76 g
NaI	2.1	4.9	4.4 ml
Glassmilk	15μl	10μl	10μl
Save TE	14.5	9.5	9.5μl
PhiX μg		0.25/0.5/0.75/1/1μl	0.5μl



#420 MF17-2 : 0.2kb fragment

80 ng/μl

880106

Nick translation to screen Tova's Library

#420 MF17-2 EcoRI 0.266 (80 ng/μl)

DNA	1.3 μl 104 ng
SalI	10
II	5
32P	20
ddH2O	13.7
total	50 μl

filter	20
HB	100 ml
SS DNA	400 μl

SAM NO	POS	TIME MIN	32P CPM	%ERROR
1	1-1	1.00	1258039.0	0.18

$1.2 \times 10^6 \times 10^2$ / 104 ng

$(1.15 \times 10^9$ /mg)

Mini-Preparation of Plasmid DNA: DNA Clones TR1 through 6 and TR8

#[redacted]42 - #448 p[redacted]EV7 / TR 1-8

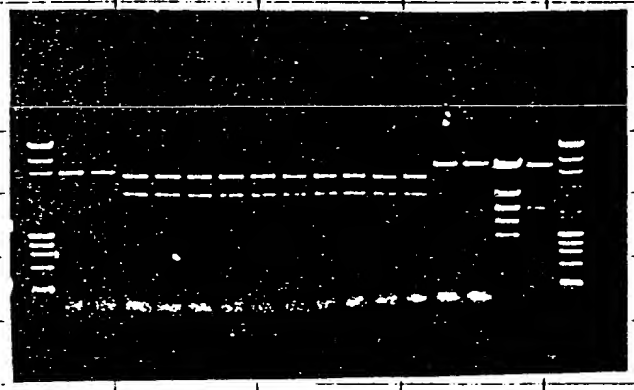
880115

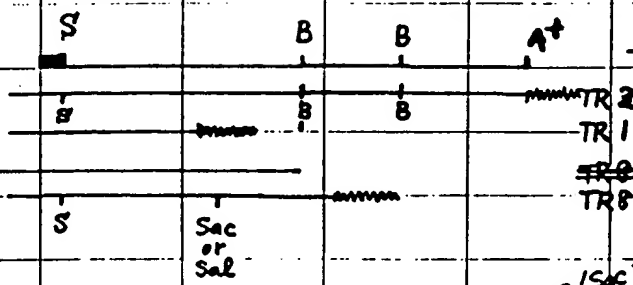
Minipreparation of plasmid DNA pCEV9 TR1-8.

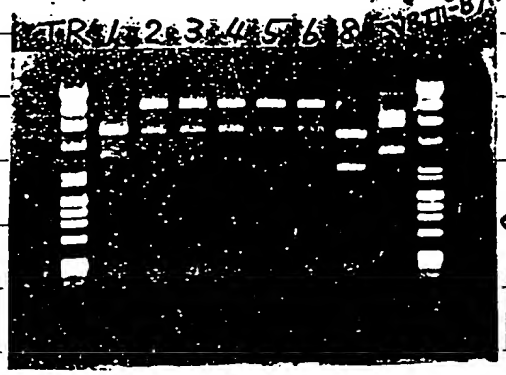
w/o RNase

DNA	5μl		5μl			Control
BamHI	1μl	SacI	1μl			SVB/TII-A/BamHI
10×B	1.5μl	10×B	1.5			SVB/BamHI XhoI
RNase	1μl		1			Control
ddH2O	6.5μl		6.5			SVB/TII-B/SacI #[redacted]42/ 78ng/λ
Total	15μl		16	37°C	30min	↓ 3.2, 0.3, 7.3, (6.5) kb
tube			SalI	1		
1	TR1	#442-1	10×B	2		BamHI
2	"	-2	ddH2O	2		
3	TR2	#443-1		20		
4	"	-2				
5	TR3	#444-1				
6	"	-2				
7	TR4	#445-1				
8	"	-2				
9	TR5	#446-1				
10	"	-2				
11	TR6	#447-1				
12	"	-2				
13	TR8	#448-1				
14	"	-2				
15	SVB/TIIA/BamHI					
16	SVB/Bam XhoI					

1/29 RNase (+)





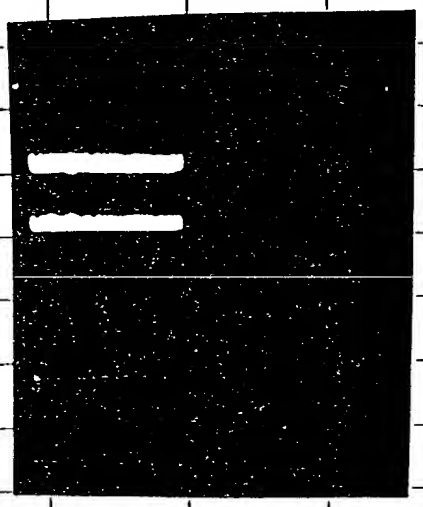


Subcloning TR4

880216

Subcloning TR4 BamHI 3.4kb fragment into SVX LTR2

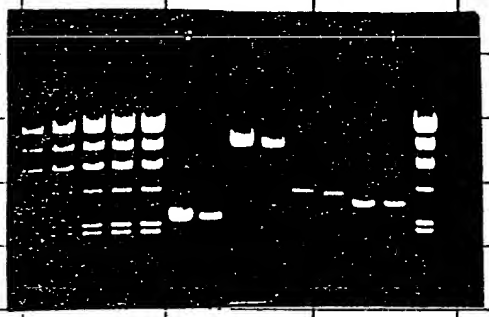
	#452	pCEV4/TR4	360μg/ml	pSVX 3032N	253μg/ml
DNA	56μl	20μg	DNA	79	20μg
BamHI	8	25u/λ	BamHI	8	
10×B	30		10×B	40	
ddH2O	206μl		ddH2O	273	
Total	300μl	check 8μl 0.8% agarose	Total	400	37°c 1hr check 8μl
			o/n	phenol ext	



cut →

TR4 SVX (BamHI)

see Reverse side



glassmilk	15μl								
	ligation	Reaction		15:00 - (3hr)					
Vector	SVX/BamHI #487	0.8	LTR2/BamHI #950 100ng/λ	0.5	pUC18/SmaI 50ng/λ #486	25ng 0.5	pUC18/SmaI	25ng 0.5	
Insert	TR4 BamHI #488 (100ng)	2	TR4 BamHI #488 (100ng)	2	#470XbaN-L ≃25ng/λ 80ng	32ng 3.2	#470XbaN-S 80ng	32ng 3.2	
Tuligase		1		1		1		1	
5×B		2		2		2		2	
ddH2O		4.2		4.5		3.3		3.3	
total		10		10		10		10	
For transformation of JM101		1μl (1/10μl)		1μl (1/10μl)		1 (3/10)		1 (3/10)	

Minipreparation of Plasmid DNA: pSSV/TR4 (α-PDGF-R) and HPR (ß-PDGF-R)

#557 #556

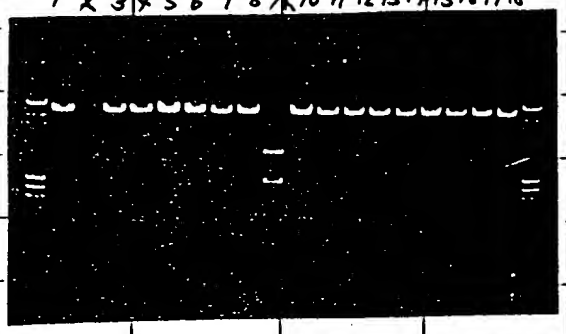
880325

Minipreparation of Plasmid DNA pSSV / TR4 or HPR

DNA	5μl
SalI	2μl
10×B	1.5
ddH2O	5.5
RNase	1
	15

#557 HPR
#556 TR4

1 X 3 4 5 6 7 8 X 10 11 12 13 14 15 16 17 18



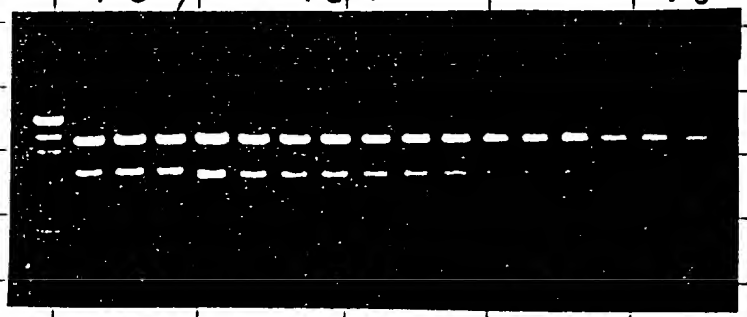
DNA	5μl
XbaI	2μl
10×B	1.5
ddH2O	5.5
RNase	1
	15
SalI	2
10×B	1.3
	18.3μl

#556 pSSV/TR4 SalI-Bbid(XbaI)

#557 pSSV/HPR Sal-Hble(XbaI)

HPR TR4

1 3 4 5 6 7 8 10 11 12 13 14 15 16 17 18



For large prep. (1l)

~~4~~

~~7~~

Binding of ^{125}I-labeled Human PDGF to Control Mouse 3T3 cells, Control COS-1 cells and COS-1 cells transfected with T11 (α-PDGF-R) HPR (ß-PDGF-R) cDNA Expression Vectors

^{125}I - PDGF binding

^{125}I - c-sis 0.5μl / well
8μl / Total 8ml

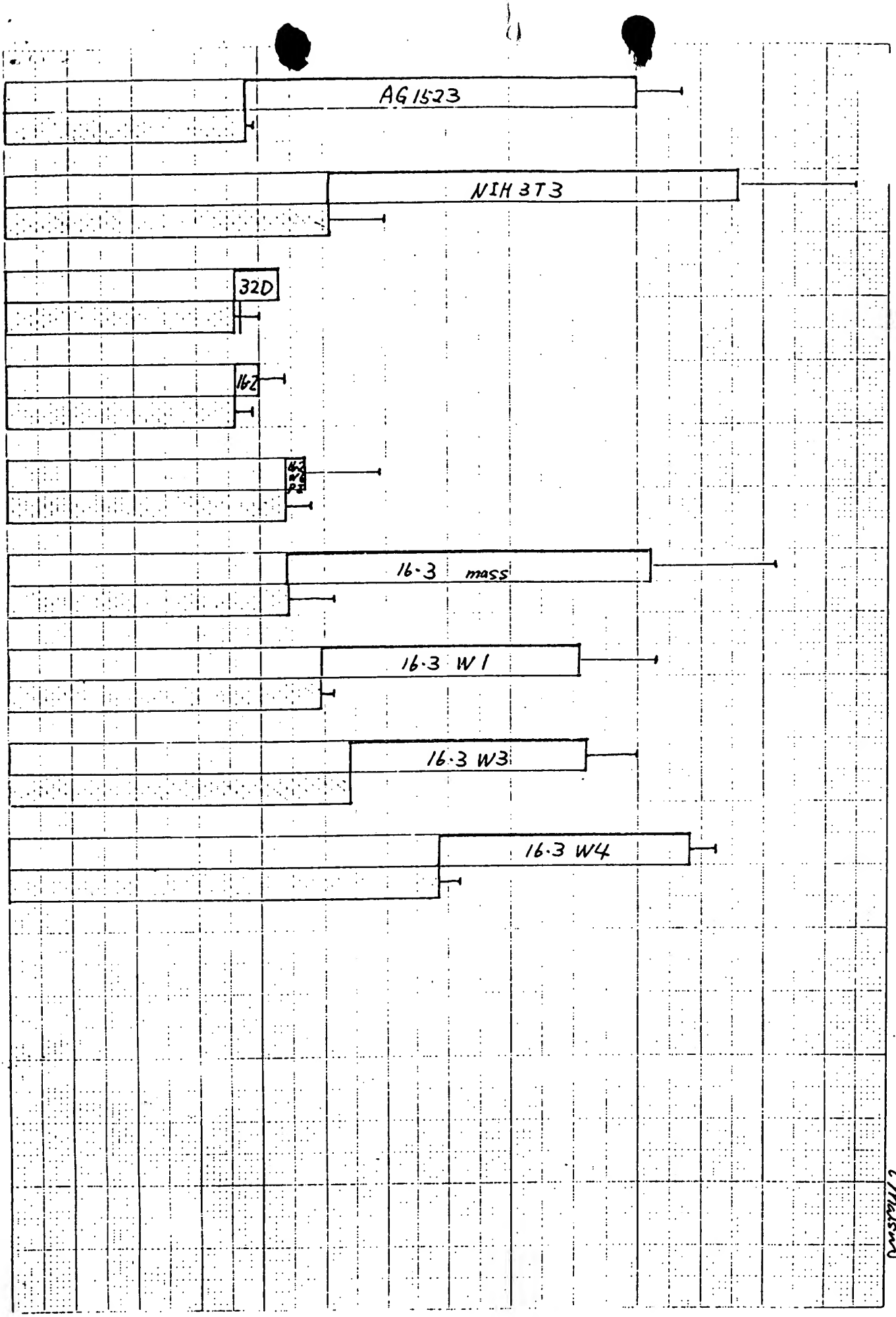
^{125}I - huPDGF 0.6μl / well (1ng)
48μl / Total 4ml

32D			8 wells +	4 wells
32D·HPR			8 wells +	4 wells
	c-sis	250 ng/μl	100%	2μl / 4 wells
	A-A	210 ng/μl	75%	2μl / 4 wells
	huPDGF	Lot 88-1198 242 ng/9.8μl		12.9μl / 4 wells
	w/o competition			
	w/o competition			(Control 1M Acetate 12.9μl
	huPDGF	242 ng/9.8μl		12.9μl / 4 wells

1. Fibronectin coating Incubate (30 min.) 2 × 12 wells / 100ng
2. Cells a) 32D JG4 85ml / flask
   b) 16.2 HPR
   cfg & resuspended in 50ml DHEM
   cfg & " 48ml DHEM
   Plate 2ml / [fibronectin coated 12 wells plate] well
   & Incubate 30 min at 37°C
3. Wash the cells & take off the non-adherant cells by using Binding Buffer (DHEM + 25mM Hepes + 1mg/ml BSA)
4. Binding RT 1hr.
   Washing by 2ml Binding Buffer × 4 times
5. Add 200μl Solbilizing Buffer & sit RT. 30min
6. Count

880328

1	32D	^{125}I-c-sis	–	306	282 ± 34					
2			–	258						
3			c-sis	256	258 ± 3					
4			"	260						
5			A-A	297	291 ± 37					
6			"	244						
7			HuPDGF	329	297 ± 46					
8			"	264						
9		^{125}I-HuPDGF	–	682	738 ± 79					
10			–	794						
11			HuPDGF	506	517 ± 15					
12			"	527						
13	32D/HPR	^{125}I-c-sis	–	2014	1997 ± 25					
14			–	1979						
15			c-sis	331	328 ± 5					
16			"	324						
17			A-A	1950	1998 ± 68					
18			"	2046						
19			HuPDGF	902	931 ± 41					
20			"	960						
21		^{125}I-HuPDGF	–	850	742 ± 153					
22			–	634						
23			HuPDGF	~~644~~ 608	576 ± 45					
24			"	~~543~~ 544						
		^{125}I-csis	20μl	943	total/well 690μl 23103					
		^{125}I-HuPDGF	20μl	1291	total/well 690μl 31630					


AG1523
NIH 3T3
32D
16-3 mass
16-3 W1
16-3 W3
16-3 W4